CLAIMS

1. A method for the purification of a cytochrome P450, wherein said method comprises:

(a) expressing in a host cell culture a cytochrome P450 molecule;

(b) recovering said cells from said culture and suspending said cells in a salt buffer having a conductivity of from 12 to 110 mS/cm;

(c) lysing said cells and removing cell debris to provide a high-salt lysate;

(d) adding to said lysate a detergent to provide a high-salt-detergent lysate; and

(e) recovering said P450 from said lysate;

provided that when said salt buffer has a concentration of from 200 to 1000mM, the P450 is not a human 2C9 P450 having position 220 substituted by proline.

2. The method of claim 1 wherein the salt buffer has a salt concentration of from 200 to 1000 mM.

3. The method of claim 1 or 2 wherein the detergent is added at 0.015 to 1.2% v/v.

4. The method of any one of claims 1 to 3 wherein step (e) is performed by:

(e(i)) binding said P450 to an affinity support;

(e(ii)) rinsing said support in a high-salt-detergent wash;

(e(iii)) removing said P450 in a high-salt-detergent buffer to provide a P450-high-salt-detergent preparation; and

(f) rapidly desalting the preparation to provide a P450-low-salt preparation.

5. The method of claim 4 wherein step (f) is performed by removing salt from said preparation by size-exclusion chromatography.

6. The method of any one of the preceding claims wherein the P450 carries a polyhistidine tag.

7. The method of any one of the preceding claims wherein the P450 is a member of the CYP1, 2, 3 or 4 family.

8. The method of claim 7 wherein the P450 is a CYP2 family member.

9. The method of claim 8 wherein the P450 is 2C9 or 2C19.

10. The method of claim any one of the preceding claims wherein the P450 comprises a deletion in its N-terminal membrane inserting element.

11. The method of claim 10 wherein the N-terminal sequence of said P450 comprises, in place of the N-terminal membrane inserting element, a sequence MAKKTSSKGR or MAYGTHSHGLFKK.

12. The method of claim 11 wherein said P450 is of SEQ ID NO:2, 4, 6 or 8.

13. The method of any one of the preceding claims which further comprises crystallizing the P450.

14. A crystal of a human cytochrome P450 selected from the group of 2C9, 2C19, 2D6 and 3A4.

15. The crystal of claim 14 wherein said P450 is 2C19 and said crystal has cell dimensions of a=158Å, b=158Å, c=212Å

(+/-5% for a, b, and c), α=90°, β=90°, γ=120°, and a space group P321.

16. The crystal of claim 14 wherein said P450 is 2D6.

17. The crystal of claim 14 wherein said P450 is 3A4 having a space group I222 and unit cell size a=77 Å, b=99 Å, c=129 Å, (+/- 5% for a, b and c), β=90°; or having a space group C2 and unit cell size a=152Å, b=101 Å, c=78Å (+/- 5% for a, b and c), α=90°, β=120°, γ=90°.

18. A method for determining the crystal structure of a cytochrome P450 which comprises preparing a crystal according to the method of claim 13, subjecting the crystal to x-ray diffraction, and analysing the diffraction pattern obtained to determine the 3-dimensional coordinates of the atoms of said P450.

19. A nucleic acid for expression of cytochrome P450 2D6 having the coding sequence for 2D6 of SEQ ID NO:5.

20. A bacterial expression vector comprising the nucleic acid of claim 19.